**DETAILED ACTION**

1. The amendment filed Mar. 1, 2011, has been entered.

2. Claims 27-30, 39-42, 46-59, 68-71, and 73-86 have been canceled. Claims 1-26, 31-38, 43-45, 60-67, and 72 are pending in this Application. Claims 4-6, 13-16, 20-26, and 60-67 are withdrawn for being directed to non-elected inventions. Claims 9-12, 19 (in part), 45 (in part) along with linking claims, claims 1-3, 7, 8, 17, 18, 31-38, 43, 44, and 72 are examined in this Office Action.

***Objections and Rejections that are Withdrawn***

3. The objection to the Oath/Declaration is withdrawn in light of the Applicant's submission of a new declaration.

4. The objection to the specification for missing sequence identifiers is withdrawn in light of the Applicant's amendments to the specification and submission of a new sequence listing.

5. The objection to the title is withdrawn in light of the Applicant's amendment to the title.

6. The objections to claims 43 and 72 are withdrawn in light of the Applicant's amendments to the claims.

7. The portion of the rejection under 35 USC 112, 2nd paragraph, that was directed to confusion about whether or not the "fragment thereof" is required to have the recited function is withdrawn in light of the Applicant's amendments to the claims.

8. The rejection of claims 1-3, 7-12, 31, 43, and 72 under 35 U.S.C. 102(b) as being anticipated by Ballicora et al is withdrawn in light of the Applicant's amendments to the claims.

***Claim Rejections - 35 USC § 112***

The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

9. Claims 1-3, 7-12, 17-19, 31-38, 43-45, and 72 remain rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point

out and distinctly claim the subject matter which applicant regards as the invention. All dependent claims are included in these rejections. The Applicant's arguments in the response filed on Mar. 1, 2011, have been fully considered but were not found to be persuasive.

The term "relative to a wild type AGP enzyme" in claim 1 is a relative term which renders the claim indefinite. The term "relative to a wild type AGP enzyme" is not defined by the claim, the specification does not provide a standard for ascertaining the requisite degree, and one of ordinary skill in the art would not be reasonably apprised of the scope of the invention. This is particularly true because the heat stability of different wild type AGP enzymes is drastically different with the AGP enzyme from potato being quite stable compared to the AGP enzyme from maize. Therefore, it is unclear which type of wild-type AGP enzyme the increased heat stability is relative to.

The Applicant argues that their amendment which replaces "relative to" with "when compared to" overcomes this rejection (see second paragraph on page 11). This is not persuasive, however, because the indefiniteness is based upon the fact that different wild-type AGP enzymes have different heat stability. For example, the potato AGP enzyme is very heat stable whereas the maize AGP enzyme is not heat stable. Therefore, it is unclear if the increased heat stability recited in claim 1 is increased compared to the wild-type maize enzyme or if it is increased compared to the wild-type potato enzyme. It is quite possible that one could make a mutant

enzyme that is more heat stable than the wild-type maize enzyme, but is less heat stable than the wild-type potato enzyme. Would this be covered by this claim?

10. Claims 1-3, 7, 9, 17-19, 31-38, 43-45, and 72 remain rejected under 35 U.S.C. 112, first paragraph, because the specification, while being enabling for a nucleic acid comprising a polynucleotide encoding a mutant small subunit with a Y36C substitution relative to SEQ ID NO:2 wherein when said mutant small subunit is expressed with a large subunit of a plant AGP enzyme to form a heterotetrameric enzyme, said heterotetrameric enzyme exhibits increased heat stability relative to the wild type maize AGP enzyme, does not reasonably provide enablement for nucleic acids encoding any mutant enzymes other than those that have a cysteine substituted for the tyrosine normally found at position 36 in the maize wild type small subunit nor does it reasonably provide enablement for mutant enzymes with increased heat stability relative to any wild type AGP enzyme other than the maize wild type enzyme nor does it reasonably provide enablement for a mutant small subunit of any heat labile plant enzyme other than the maize AGP enzyme. The specification does not enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the invention commensurate in scope with these claims. The Applicant's arguments in the response filed on Mar. 1, 2011, have been fully considered but were not found to be persuasive.

The claimed invention is not supported by an enabling disclosure taking into account the *Wands* factors. *In re Wands*, 858/F.2d 731, 8 USPQ2d 1400 (Fed. Cir. 1988). *In re Wands* lists a number of factors for determining whether or not undue experimentation would be required by one skilled in the art to make and/or use the invention. These factors are: the quantity of experimentation necessary, the amount of direction or guidance presented, the presence or absence of working examples of the invention, the nature of the invention, the state of the prior art, the relative skill of those in the art, the predictability or unpredictability of the art, and the breadth of the claim.

The claims are broadly drawn to a polynucleotide encoding a mutant small subunit of a heat labile plant ADP glucose pyrophosphorylase (AGP) enzyme, wherein when said small subunit is expressed with a large subunit of a plant AGP enzyme to form a mutant enzyme, said mutant enzyme exhibits increased heat stability relative to a wild type AGP enzyme and to plants, compositions and expression constructs comprising said polynucleotide

Applicants teach polynucleotides encoding mutant AGP small subunits having a cysteine substituted for the tyrosine that is normally found at position 36 in the wild type maize AGP small subunit (Y36C); wherein the resulting AGP enzyme has increased heat stability relative to the wild-type maize AGP enzyme (see Table 3 on page 31). This includes mutant AGP small subunits that had an

additional amino acid inserted between residues 34 and 35 of the wild-type maize small subunit in addition to the Y36C substitution.

Applicants do not teach any heat stabilized AGP subunits that did not have a cysteine substituted for the tyrosine that is normally found at position 36 in the wild type maize AGP small subunit, nor did they teach any mutants that had an increase in stability relative to non-maize wild-type AGP enzymes, nor did they teach any heat labile plant AGP enzymes other than the maize AGP enzyme.

For example, in the prior art (Plant Physiology (1995) Vol. 109; pp. 245-251), Ballicora et al teach that the N terminus of the small subunit from the potato AGP enzyme is important for its heat stability (see right column on page 248), however the increase in heat stability was relative to a truncated version of the AGP enzyme, it was not relative to the heat stability of the potato wild-type enzyme. Furthermore, in the prior art (Biochem. Biophys. Res. Comm. (1999) Vol. 257; pp. 782-786), Ballicora et al teach that the heat stability of the potato AGP is the result of di-sulfide bonds formed from a cysteine residue in the small subunit (see paragraph bridging pages 783-784). For this reason, mutations that do not add a cysteine residue to facilitate formation of di-sulfide bonds are highly unlikely to increase the heat stability of the maize heat labile AGP enzyme.

Given the lack of guidance in the instant specification, undue trial and error experimentation would be required for one of skill in the art to identify heat labile AGP enzymes other than the maize wild-type enzyme or for one of skill in the art to

identify mutations, other than addition of cysteine residues in the N-terminus of the small subunit of the maize AGP enzyme that would result in an increase in heat stability of the enzyme. It would also require undue trial and error experimentation for one of skill in the art to identify mutations that would result in an increase in heat stability relative to wild-type potato AGP which is already quite heat stable.

Therefore, given the breadth of the claims; the lack of guidance and working examples; the unpredictability in the art; and the state-of-the-art as discussed above, undue experimentation would be required to make and use the claimed invention, and therefore, the invention is not enabled throughout the broad scope of the claims.

The Applicant argues that the specification has provided guidance about making mutations, and there was quite a bit known in the art about the small subunit of AGP from different plants, and that experimentation to determine if a mutant has increased heat stability is routine experimentation (see pages 11-12 of the response).

The Examiner agrees with these statements, and therefore, the Examiner agrees that one of skill in the art would be able to add cysteines to the N-terminal region or substitute cysteines for other amino acids that are in the N-terminal region of an AGP small subunit that is known to be heat labile, and then it would be routine experimentation to determine if the modified AGP small subunit is more heat stable than the wild-type subunit from which it was derived. However, the

instant claims recite "increased heat stability when compared to a wild type AGP enzyme", which encompasses being compared to wild-type potato AGP which is already very heat stable (see rejection under 35 USC 112, 2nd paragraph, above). The instant claims do not require that the increase in heat stability is relative to the particular heat labile wild-type AGP subunit that was modified by a mutation in its N-terminal portion. Furthermore, many of the instant claims do not require a cysteine to be added, and it appears that the specification would only be enabling for mutations that insert a cysteine or substitute a cysteine for another residue, because the heat stability is the result of di-sulfide bonds that require a cysteine residue to be added.

11. Claims 1-3, 7, 9, 17-19, 31-38, 43-45, and 72 remain rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention. The Applicant's arguments in the response filed on Mar. 1, 2011, have been fully considered but were not found to be persuasive.

The claims are broadly drawn to a polynucleotide encoding a mutant small subunit of a heat labile plant ADP glucose pyrophosphorylase (AGP) enzyme, wherein when said small subunit is expressed with a large subunit of a plant AGP

enzyme to form a mutant enzyme, said mutant enzyme exhibits increased heat stability relative to a wild type AGP enzyme and to plants, compositions and expression constructs comprising said polynucleotide

Applicants describe polynucleotides encoding mutant AGP small subunits having a cysteine substituted for the tyrosine that is normally found at position 36 in the wild type maize AGP small subunit (Y36C); wherein the resulting AGP enzyme has increased heat stability relative to the wild-type maize AGP enzyme (see Table 3 on page 31). This includes mutant AGP small subunits that had an additional amino acid inserted between residues 34 and 35 of the wild-type maize small subunit in addition to the Y36C substitution.

Applicants do not describe any heat stabilized AGP subunits that did not have a cysteine substituted for the tyrosine that is normally found at position 36 in the wild type maize AGP small subunit, nor did they describe any mutants that had an increase in stability relative to non-maize wild-type AGP enzymes, nor did they describe any heat labile plant AGP enzymes other than the maize AGP enzyme.

The essential features of the instant invention are that mutant enzyme comes from a heat labile plant AGP, and that the mutant enzyme has an increase in heat stability relative to a wild type AGP enzyme (see claim 1).

In the prior art (Plant Physiology (1995) Vol. 109; pp. 245-251), Ballicora et al teach that the N terminus of the small subunit from the potato AGP enzyme is important for its heat stability (see right column on page 248), however the increase

in heat stability was relative to a truncated version of the AGP enzyme, it was not relative to the heat stability of the potato wild-type enzyme. Furthermore, in the prior art (Biochem. Biophys. Res. Comm. (1999) Vol. 257; pp. 782-786), Ballicora et al teach that the heat stability of the potato AGP is the result of di-sulfide bonds formed from a cysteine residue in the small subunit (see paragraph bridging pages 783-784). For this reason, mutations that do not add a cysteine residue to facilitate formation of di-sulfide bonds are highly unlikely to increase the heat stability of the maize heat labile AGP enzyme.

The Federal Circuit has recently clarified the application of the written description requirement to inventions in the field of biotechnology. The court stated that, "A description of a genus of cDNAs may be achieved by means of a recitation of a representative number of cDNAs, defined by nucleotide sequence, falling within the scope of the genus or of a recitation of structural features common to members of the genus, which features constitute a substantial portion of the genus." See *University of California v. Eli Lilly and Co.*, 119 F. 3d 1559; 43 USPQ2d 1398, 1406 (Fed. Cir. 1997).

The Applicants fail to describe a representative number of heat labile plant AGP enzymes. The Applicants only describe the maize AGP enzyme as being heat labile. The Applicants fail to describe a representative number of mutant small subunits that result in an increase in stability relative to any wild-type AGP enzyme. The Applicants only describe mutants with increased stability relative to

the wild-type maize AGP enzyme. The Applicants fail to describe a representative number of mutant small subunits with mutations across the N-terminal portion of the small subunit, wherein the mutations result in an increase in heat stability. The Applicants only describe mutants that have a cysteine substituted for the tyrosine that is normally found at position 36 in the wild type maize AGP small subunit. Furthermore, the Applicants fail to describe structural features common to members of the claimed genus of heat-stabilized small subunits. Hence, Applicants fail to meet either prong of the two-prong test set forth by *Eli Lilly*. Furthermore, given the lack of description of the necessary elements essential for increasing the heat stability of the AGP enzyme, it remains unclear what features identify mutant small subunits capable of such activity. Since the genus of small subunits with mutations in the N-terminus that increase the heat stability of the enzyme has not been described by specific structural features, the specification fails to provide an adequate written description to support the breadth of the claims.

Nucleic acids that encode mutant small subunits of AGP encompass multitudes of molecules, many of which would not encode heat-stabilized AGP enzymes when co-expressed with an AGP large subunit in a plant cell, and most of which were not in the Applicant's possession at the time of filing. The Applicants have reduced to practice only three mutants that demonstrate increased heat stability relative to wild-type maize AGP, all of which have a cysteine substituted for the tyrosine that normally appears at position 36 in the wild-type maize small

subunit. Accordingly, the specification fails to provide an adequate written description to support the genus of nucleic acids encoding mutant small subunits of heat labile AGP enzymes that have increased heat stability relative to wild-type AGP enzymes as set forth in the claims. (See Written Description guidelines published in 2008 online at http://www.uspto.gov/web/menu/written.pdf).

The Applicant argues that the specification has provided guidance about making mutations, and there was quite a bit known in the art about the small subunit of AGP from different plants, and that experimentation to determine if a mutant has increased heat stability is routine experimentation (see pages 11-12 of the response). The arguments are more applicable to the lack of enablement rejection, rather than the written description rejection, but they were set forth in response to the written description rejection as well.

The Examiner agrees with these statements, and therefore, the Examiner agrees that one of skill in the art would be able to add cysteines to the N-terminal region or substitute cysteines for other amino acids that are in the N-terminal region of an AGP small subunit that is known to be heat labile, and then it would be routine experimentation to determine if the modified AGP small subunit is more heat stable than the wild-type subunit from which it was derived. However, the instant claims recite "increased heat stability when compared to a wild type AGP enzyme", which encompasses being compared to wild-type potato AGP which is already very heat stable (see rejection under 35 USC 112, 2nd paragraph, above).

The instant claims do not require that the increase in heat stability is relative to the particular heat labile wild-type AGP subunit that was modified by a mutation in its N-terminal portion. The instant application has not described any mutant enzymes that are more heat stable than the wild-type potato enzyme.

Furthermore, many of the instant claims do not require a cysteine to be added, and the instant specification has only reduced to practice mutants that substitute a cysteine for another residue. Given that there are no embodiments reduced to practice that do not include the addition of a cysteine residue, and given that the art teaches that the heat stability is the result of di-sulfide bonds that require a cysteine residue to be added, the instant specification has not provided an adequate description of any mutants that do not have additional cysteines at the N-terminal region, nor has the instant specification provided an adequate description of mutants that have greater heat stability than wild-type potato AGP enzyme.

***Claim Rejections - 35 USC § 103***

The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

12. Claims 1, 2, 17-19, 31-38, 43-45, and 72 remain rejected and amended claims 3 and 7-12 are rejected under 35 U.S.C. 103(a) as being unpatentable over Greene et al (PNAS (1998) Vol. 95; pp. 13342-13347) in view of Giroux, M. (US Pre-Grant Publication US 2003/0150027; for application no. 10/116,868, filed on April 5, 2002, with priority to 09/516,250, filed on Mar. 1, 2000) and further in view of Ballicora et al (Biochem. Biophys. Res. Comm. (1999) Vol. 257; pp. 782-786). The Applicant's arguments in the response filed on Mar. 1, 2011, have been fully considered but were not found to be persuasive.

The claims are drawn to a polynucleotide encoding a mutant small subunit of a heat labile plant ADP glucose pyrophosphorylase (AGP) enzyme, wherein when said small subunit is expressed with a large subunit of a plant AGP enzyme to form a mutant enzyme, said mutant enzyme exhibits increased heat stability relative to a wild type AGP enzyme and to plants, compositions and expression constructs comprising said polynucleotide.

Greene et al teach mutations in the large subunit of the maize AGP enzyme that result in enhanced heat stability (see entire article). They specifically teach one that is referred to as HS33 (see left column on page 13344). They teach a comparison of large subunit sequences from maize, wheat, barley, rice, and potato (see Figure 2 on page 13344).

Green et al do not teach mutations in the small subunit of a heat labile AGP enzyme. Nor do they teach transgenic plants expressing mutant small subunits or a polynucleotide that encodes both a small subunit and a large subunit.

Giroux teaches transgenic plants expressing a mutant form of AGP (see entire document) including both monocots (see claim 41) and dicots (see claim 45); with specific suggestions for wheat (see examples 1 and 2 on pages 15 and 16), rice (see paragraph 0220 on page 17), and pea (see paragraph 0221 on page 17).

Ballicora et al teach that a cysteine at the N-terminus of the potato AGP small subunit is important for the heat stability of the enzyme and that di-sulfide bonds formed from a cysteine residue in the small subunit are important for this stability (see paragraph bridging pages 783-784).

At the time the invention was made, it would have been obvious and within the scope of one of ordinary skill in the art to make mutations in the maize AGP small subunit to introduce cysteine residues in the N-terminus with the goal of providing the necessary SH group for forming di-sulfide bonds to increase the heat stability of the enzyme. Given the teachings of Giroux that AGP is the rate limiting step in starch biosynthesis in plants (see paragraph 0011 on page 1), and the teachings of Giroux demonstrating that increasing the amount of AGP provides an increase in seed production and biomass production (see paragraph 0022 on page 1; and see experimental results in Table 1 on page 16), one would have been motivated to increase the amount of AGP in a transgenic plant. Given the teaching of

Ballicora et al that di-sulfide bonds via cysteine residues in the N-terminal region of the small subunit can result in an increase in heat stability, one would have been motivated to substitute cysteine residues or to insert cysteine residues into the maize small subunit with a goal of increasing the heat stability of the AGP. Given the success of Greene et al in expressing mutant forms of the AGP large subunit that have increased heat stability, and given the success of Ballicora et al in producing transgenic plants with increased seed production and biomass production by increasing the amount of AGP, one would have had a reasonable expectation of success in expressing mutant maize AGP small subunits with increased heat stability that would result in an increase in seed production and biomass.

The additional limitations of co-expressing both the mutant small subunit and a large subunit from the same polynucleotide are obvious variations of what was taught in the art. The experiments by Ballicora et al utilize two separate plasmids to express the small subunit and large subunit; however, bi-cistronic expression vectors for co-expression of two polypeptides in plant systems were well known in the art at the time of filing (for example, co-expression of the heavy chain and light chain of an antibody; or for example, expression of a protein of interest in addition to a selectable marker). For this reason, a polynucleotide that encodes both the mutant small subunit and the large subunit is an obvious variation.

The Applicant argues that Greene et al and Giroux are directed to mutations in the large subunit of AGP and do not teach or suggest mutations in the small

subunit; and the Applicant argues that Ballicora et al teaches mutations in the small subunit of the potato AGP which is not heat labile as required by the instant claims (see second paragraph on page 13 of the response). This is not persuasive, however, because it is arguing against the references individually rather than considering the combined teachings as a whole. In response to applicant's arguments against the references individually, one cannot show nonobviousness by attacking references individually where the rejections are based on combinations of references. See *In re Keller*, 642 F.2d 413, 208 USPQ 871 (CCPA 1981); *In re Merck & Co.*, 800 F.2d 1091, 231 USPQ 375 (Fed. Cir. 1986). The heat labile maize enzyme is taught by Greene et al and Giroux, and the small subunit and requirement for cysteines in the N-terminus of the small subunit to confer heat stability is taught by Ballicora et al.

The Applicant argues that there is not suggestion or reasonable expectation of success found in the cited references (see paragraph bridging pages 13-14 of the response). This is not persuasive, however, because Giroux provides a motivation for expressing heat stable AGP in plants, and Ballicora et al provide the suggestion that adding cysteines to the N-terminus of the small subunit of AGP could result in di-sulfide bonds that would increase heat stability. These teachings are the basis for combining the references to arrive at the claimed invention. Given that Giroux was successful at expressing AGP with increased heat stability, and given the teachings of Ballicora et al that would suggest adding cysteines to the N-terminus

of the small subunit of AGP to arrive at increased heat stability, one would have had a reasonable expectation of success to produce an AGP enzyme with increased heat stability relative to the maize AGP enzyme by adding cysteines to the N-terminus of the small subunit.

13. No claim is allowed.

***Allowable Subject Matter***

14. The Examiner suggests the following claim language: - - A nucleic acid comprising a polynucleotide encoding a mutant small subunit of an ADP glucose pyrophosphorylase (AGP) enzyme, wherein said mutant small subunit comprises a substitution of a cysteine for the tyrosine at position 36 of the wild-type maize small subunit, and wherein said mutant small subunit optionally further comprises an insertion of an amino acid between the serine at position 34 and the threonine at position 35 of the wild-type maize small subunit. - -

Although the prior art suggests mutations that would substitute or add cysteine residues at the N-terminus of the maize AGP small subunit (see rejection under 35 USC 103, above), the prior art does not teach or suggest a substitution, specifically, of a cysteine for the tyrosine at position 36 of the wild-type maize AGP small subunit.

It is the Examiner's position that the recommended claim language, above, would be free of all of the rejections of record. If the Applicant confirms that SEQ ID NOs: 4, 8, and 10 each are mutant subunits with a cysteine for the tyrosine at position 36 of the wild-type maize small subunit, and optionally an insertion of an amino acid between the serine at position 34 and the threonine at position 35 of the wild-type maize small subunit, then all of these sequences could be rejoined for comprising a shared special technical feature, and for being "linked" by an allowable claim (ie. the recommended claim language).

15. Applicant's amendment necessitated the new ground(s) of rejection presented in this Office action. Accordingly, **THIS ACTION IS MADE FINAL**. See MPEP § 706.07(a). Applicant is reminded of the extension of time policy as set forth in 37 CFR 1.136(a).

A shortened statutory period for reply to this final action is set to expire THREE MONTHS from the mailing date of this action. In the event a first reply is filed within TWO MONTHS of the mailing date of this final action and the advisory action is not mailed until after the end of the THREE-MONTH shortened statutory period, then the shortened statutory period will expire on the date the advisory action is mailed, and any extension fee pursuant to 37 CFR 1.136(a) will be calculated from the mailing date of the advisory action. In no event, however, will

the statutory period for reply expire later than SIX MONTHS from the date of this final action.

16. Any inquiry concerning this communication or earlier communications from the examiner should be directed to CATHY K. WORLEY whose telephone number is (571)272-8784. The examiner is on a variable schedule but can normally be reached on M-F 10:00 - 4:00, with additional variable hours before 10:00 and after 4:00 with additional variable hours before 10:00 and after 4:00.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Anne Marie Grunberg, can be reached on (571) 272-0975. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free).

/Cathy K. Worley/
Primary Examiner, Art Unit 1638